ACTIVITY OF SOME SERUM ENZYMES IN EXPERIMENTAL RENAL ISCHEMIA

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Enzymological investigations are widely used in present-day clinical practice in the diagnosis of various diseases, including those associated with necrotic changes in the tissues based on hypoxia (myocardial infarction). According to the prevailing opinion, enzymes are "flushed out" of the necrotic foci into the blood [3]. Adherents of a different view consider that the increased serum enzyme concentration is one component of a nonspecific "acute syndrome", characteristic of many diseases [6]. It is not impossible that the increase may result from both mechanisms [5].

In experiments on cats we studied the effect of a temporary occlusion of the renal circulation on the concentration of transaminase, aldolase, alkaline phosphatase, and copper-oxidase in the blood.

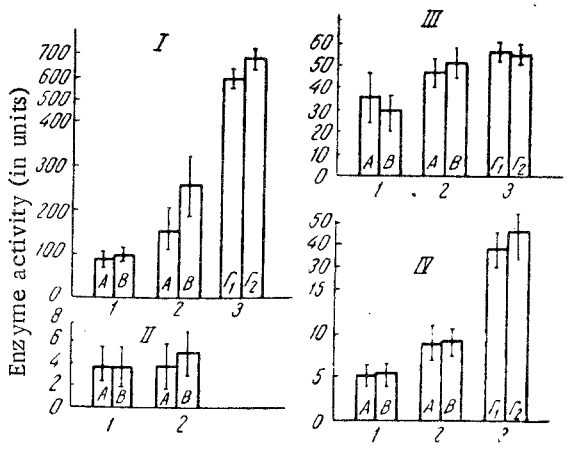
EXPERIMENTAL METHOD

Animals were anesthetized with hexobarbital (60-80 mg/kg) or thiopental (60-90 mg/kg), which was injected intraperitoneally in the form of a freshly prepared solution. The left kidney was approached by laparotomy and a cannula was introduced via the testicular vein into the renal vein. If the testicular vein was of too narrow a caliber, the cannula was introduced into the inferior vena cava so that its end was at the level of entry of the renal vein. A second cannula was introduced against the blood flow into the abdominal aorta below the origin of the renal arteries. Before operation each cat received an intravenous injection of heparin (1000-1500 units).

The experiment began with the taking of control samples of arterial blood and venous blood flowing from the kidney. During taking of the samples of venous blood reflux was prevented by clamping the renal vein proximally to the site of introduction of the cannula. A Dieffenbach's clamp was then applied to the left renal artery, thereby

TABLE 1. Activity of Blood Enzymes at the Beginning of the Experiment and 1 and 3 h after Clamping the Renal Artery

Enzyme	Before clamping the renal artery	After clamping the renal artery	
		after 1 h	after 3 h
GOT			
Arterial blood	97 (77–117)	101 (81–121)	99 (89–100)
Venous blood	101 (78–124)	109 (69–149)	108 (93–123)
Aldolase			
Arterial blood	28 (23–33)	29 (25–33)	32 (26–38)
Venous blood	27 (22–32)	31 (23–39)	33 (27–39)
Alkaline phosphatase			
Arterial blood	6.0 (5.2–6.8)	6.8 (5.4–8.2)	7.1 (5.7–8.5)
Venous blood	6.2 (4.9–7.5)	8.0 (6.9–9.1)	7.5 (6.5–8.5)
Copper-oxidase			
Arterial blood	5.9 (4.7–7.1)	4.9 (2.8–7.0)	3.3 (2.0–4.6)
Venous blood	6.2 (2.4–10)	7.4 (5.4–9.4)	6.3 (3.7–8.9)



Activity of GOT (I), copper-oxidase (II), aldolase (III), and alkaline phosphatase in the serum of arterial (A) and venous (B) blood before clamping (1) and 6 h after clamping (2) the renal artery, and also in homogenates (3) of the ischemic (H_1) and intact kidney (H_2).

TABLE 2. Activity of the Serum Enzymes after Laparotomy

Enzyme	Immediately after laparotomy	6 hours after laparaotomy
GOT		
Arterial blood	80 (66–94)	85 (63–107)
Venous blood	72 (54–90)	75 (56–94)
Aldolase		
Arterial blood	23 (17–29)	30 (22–38)
Venous blood	25 (15–35)	33 (21–45)
Alkaline phosphatase		
Arterial blood	10 (5.6–14.4)	10.6 (7.1–14.1)
Venous blood	9.6 (7–12.2)	10.6 (6.3–14.9)

excluding the kidney from the general circulation. After definite intervals of time blood samples were again taken for estimation of the enzyme activity. When samples of venous were taken the clamp was removed from the artery.

In ten experiments blood samples were taken 1 and 3 h after clamping the renal artery, and in 12 experiments 6 h after exclusion of the kidney from the circulation. The animal was then sacrificed, and homogenates (1:10) prepared from both kidneys, in which the enzyme activity was also investigated.

The activity of the serum glutamino-oxalacetic transaminase (GOT) was determined by Paskhina's method [2], aldolase by the method described by A. V. Anan'ev [1], alkaline phosphatase by Bodansky's method [4], and copper-oxidase by Ravin's method [7]. The experimental results concerning the enzyme activity were analyzed statistically, with calculation of the arithmetical mean values and their confidence limits ($P = 0.05$).

EXPERIMENTAL RESULTS

It will be clear from Table 1 that the GOT and aldolase activity in both the arterial and venous blood was essentially unchanged. The difference in alkaline phosphtatase activity also was not statistically significant ($P > 0.05$).

The decrease in the copper-oxidase activity observed in the arterial blood 3 h after occlusion of the renal blood flow ($P < 0.05$) cannot be satisfactorily explained. Hence, severe renal ischemia for 3 h was not accompanied by an increase in the activity of the serum enzymes. This applied both to the enzymes contained in the kidney tissue (GOT, aldolase, alkaline phosphatase) and to the enzymes not present in the kidney tissue (copper-oxidase).

The figure shows that after clamping the renal artery for 6 h the activity of GOT ($P < 0.02$), aldolase ($0.1 > P > 0.05$), and alkaline phosphatase ($P < 0.01$) increased significantly. Moreover, the GOT activity of the outflowing venous blood was significantly higher than in the arterial blood ($P < 0.05$). In the homogenates of ischemic renal tissue a decrease in GOT activity was observed ($P < 0.02$), while the aldolase and alkaline phosphatase activity was unchanged.

In order to ascertain whether the change in the activity of the serum enzymes was connected with the operation (laparotomy), control experiments not including clamping the renal artery were performed on six cats. The activity of the enzymes was determined in the arterial and venous blood taken immediately after laparotomy and 6 h after the operation.

It will be seen in Table 2 that the activity of GOT, alkaline phosphatase, and copper-oxidase was essentially unchanged. The increase in the mean values of the aldolase activity also was not statistically significant. Hence, the changes in the activity of the enzymes in the blood observed after ischemia for 6 h were not connected with the laparotomy.

The absence of any difference in the alkaline phosphatase and aldolase activity in the arterial blood and the venous blood from the kidney after ischemia for a period of 6 h demonstrates that the ischemic kidney is not the im-

mediate source of the increased activity of these enzymes in the arterial blood. So far as the GOT is concerned, on the one hand the increase in the activity of this enzyme after exclusion of the renal circulation is evidence of the existence of an extrarenal mechanism of the increase in its concentration in the arterial blood. On the other hand, the fact that the GOT activity in the venous blood flowing from the kidney was higher than in the arterial blood suggests that this enzyme is also "flushed out" of the ischemic kidney.

SUMMARY

Experiments were staged on cats to analyze the mechanism of hyperenzymemia in ischemic injury of the organ. A study was made of the effect produced by temporary arrest of renal circulation on the content of transaminase, aldolase, alkaline phosphtase and copper-oxidase in the arterial and venous (flowing from the kidney) blood. As established, hyperenzymemia following a 6-hour renal ischemia may be caused by the passing of enzymes into the blood from other organs, as well as by "washing out" of some enzymes from the ischemic kidney.

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